



COPY OF PAPERS
ORIGINALLY FILED

LABORATORY NOTEBOOK

MBX-1212

Notebook No.: 6

Assigned to: Lara Madison

Date: _____

Use Nalge Cat. No.

6301-1000
to reorder.

Copyright 1973, Nalge Company
Printed in U.S.A.

BEST AVAILABLE COPY



Design primers for sequencing AI/B1 Fusions

1296..2036 ="phbB"

20
1296
740

FB.1

FB.2

FB.3

atgac tcagcgcat gcgtatgtga
ccggcgcat ggggtgtatc ggaaccgcca tttgccagcg gctggccaag gatggcttc
gtgtggtggc cgggtgcggc cccaactcgc cgcgcgcga aaagtggctg gacgagcaga
aggccctggg cttcgatttc attgcctcgg aaggcaatgt ggtgactgg gactcgacca
agaccgcatt cgacaaggte aagtcgagg tcggcgaggt tgggtgctg atcaacaacg
ccggtatcac ccgcgacgtg gtgttcgca agatgaccgc cgcgactgg gatcggtga
tcgacacca cctgactctg ctgttcaacg tcaccaagca ggtgatcgac ggcattggcg
accgtggctg gggccgcac gtcaacatct cgtcggtgaa cgggcagaag ggcagttcg
gccagacca ctactccacc gccaaaggcg gctgcatgg cttcaccatg gactggcgc
aggaagtggc gaccaaggcg gtgaccgtca acacggctc tcggggctat atcgccaccg
acatgggtcaa ggcgactcgc caggacgtgc tcgacaagat gtcgcgacg atcccggtca
agcgctggg cctgccgaa gagatcgct cgatctgcgc ttggtgtcg tcggaggagt
ccggtttctc gaccggcgcc gacttctgc tcaacggcgg cctgcatatg ggctga

Table of Contents

40..1221 ="phbA"

FA.1

FA.2

FA.3

FA.4

a tgactgactg tgtcatcgta
tcgcgcgcc gcaccgcgg cggaagttt ggcggctcgc tggccaagat ccggcaccg
gaactgggtg cctgtgcat caaggccgcg ctggagcgcg ccggcgtaa gccggagcag
gtgagcgaag tcatcatggg ccagggtctg accgcgggtt cgggcagaa cccgcacgc
caggccgcga tcaaggccgg cctgcggcg atggtgccc ccatgaccat caacaagggt
tgccgctcgg gctgaaggc cgtgatctg gccgccaacg cgatcatggc gggcgacgc
gagatcgtg tggccgggg ccaggaaac atgagcgcg ccccgacgt gctgccggg
tcgcgcgatg gtttccgat gggcgatgcc aagctggctg acaccatgat cgtcgacgc
ctgtgggacg tgtacaacca gtaccacatg ggcacacgc cggagaacgt ggccaaggaa
tacggcatca cagcgaggc gcaggatgag ttgcgcgtg gctcgagaa caaggccgaa
gccgcgcga aggcggcga gtttgacgaa gagatcgcc ggtgctgat ccgcagcgc
aaggcgacg cgggtggcct caagaccgac gagttcgtc gccaggcgc cagctggac
agcatgtcgc gectcaagcc cgccttcgac aaggccgga cggtgaccg ggccaagcc
tcgggctga acgacggcgc cgcgcgggt gtggtgatgt cggcgccaa ggccaaggaa
ctgggctga ccccgctggc cagcatcaag agctatgcca acgcgggtg cgateccaag
gtgatgggca tgggcccgtt gccggcctc aagcgcgcc tgcgcgcgc cgagtggac
ccgcaagacc tggacctgat agagatcaa gaggccttg ccgcagagc gctggcggtg
eatcagcaga tgggctggga cactccaag gtcaatgta acggcggcgc catcgccatc
ggccacccga tcggcgctc gggctgccgt atcctggtga cgctgctga cgagatgaag
cgccgtgacg cgaagaagg cctggcctc ctgtgcatc gggcgccat gggcggtggc
ctggcagtcg agcgcaata a

BEST AVAILABLE COPY

Continued on Page

17

Read and Understood By

Lucia M. Melanson

Signed

Date

[Signature]

Signed

Date

AB FUSION CHARACTERIZATION

noticed I had left the MBx240 pTrc AB fusions on the bench for approximately a week.

MBx240 pTrc AB # 10, 11, 12, 13

LB Amp

clear / tan

no granules

LB Amp

white

granules

1% glucose

LB Amp

white

granules

1% glucose

0.1mM IPTG

eye

light microscopy

start LB glucose O/N culture from freezer stock

50% x = 1% 100ml

2ln + 0.5ln

1/12-16-97 dilute cultures 1:100 into LB 1% glucose Amp

O/N

@ time dil

4 hrs post -

3ln after inoc.

induction

pTrc-AB 10	0.46335	4.63	0.7839	1.344	1.0204
11	0.55087	5.51	0.7959	1.324	1.0027
12	0.42090	4.21	0.7931	1.513	0.9694
13	0.50844	5.84	0.7476	1.268	0.9897

induced

⊕

⊖

5mM IPTG

take 300ul

Samples → 4°C

BEST AVAILABLE COPY

Continued on Page

92

Read and Understood By

Signed

Date

Signed

Date

Cont. from p. 88

LOOK @ CULTURES UNDER MICROSCOPE.

DID NOT HAVE TIME WHEN I TOOK
SAMPLES FOR WESTERN.

THEREFORE SAMPLES HAVE BEEN @ RT FROM 5 PM → 11:15 AM
* SAMPLES ARE IN LB 1% GLUCOSE *

+ IPTG -

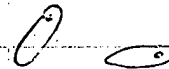
PTC LAB #10

granules
look like peas in
a pod

single granules @ poles



very small



#11

"

"

#12

"

"

#13

"

"

→ many cells w/ inclusion bodies

Majority of cells in presence 5 mM IPTG
are 2-3x ~~more~~ normal length
while the majority of cells without IPTG
are 1x cell lengths.

of course there is always a small majority
that are 5-10x cell length but only in
IPTG samples

Continued on Page

94

BEST AVAILABLE COPY

Read and Understood By

Signed

Date

Signed

Date